A Phylogenetic Classification of Ophioglossaceae

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Abstract

A cladistic approach was undertaken to construct a phylogenetic classification of the Ophioglossaceae. The characters used are: rhizome, stele, leaf sheath, size and cutting of trophophyll, vascular branching in leaf axes, venation, position of sporophyll on trophophyll, form and construction of sporophyll, position and dehiscence of sporangia, and suspensor. Character state, primitive (plesiomorphic) vs. advanced (apomorphic), was estimated for each character. Based on presumed character phylogenies, a cladogram representing phylogenetic relationships was drawn. Although in previous classifications (e.g. Clausen, 1938) the Ophioglossaceae were classified into three genera, *Botrychium, Helminthostachys* and *Ophioglossum,* it is contended here that the genus *Botrychium sensu* Clausen is not a monophyletic group. In this paper I propose a new classification recognizing six genera and arrange them as in Cracraft's (1974) sequence as follows: 1 (most primitive), *Botrypus;* 2, *Japanobotrychium;* 3-1, *Sceptridium;* 3-2, *Helminthostachys;* 4, *Botrychium;* 5 (most advanced), *Ophioglossum.*

The Ophioglossaceae have a combination of unique or rare morphological characters among the extant lower vascular plants: three-dimensional leaf architecture, collateral vascular bundles (sympodial vascular system; Stevenson, 1980) with secondary growth in many species, hair-lacking roots with endophytic fungi, noncircinate vernation of leaves, a soft and fleshy plant body devoid of sclerenchyma, massive eusporangia, and subterranean, mycorrhizic, massive gametophytes of fundamentally axial organization (Bierhorst, 1971; Foster and Gifford, 1974). Because of those characters the family is regarded as an isolated group composing the monotypic order Ophioglossales.

The usual systematic treatment is that the Ophioglossales, along with the Marattiales, constitute a eusporangiate group in Filicopsida or ferns (e.g. Christensen, 1938; Eames, 1936; Smith, 1955), or the order is one of three or more orders composing Filicopsida (e.g. Copeland, 1947; Foster and Gifford, 1974; Pichi Sermolli, 1973: Tryon and Tryon, 1982). However, a real relationship of the Ophioglossales with the Marattiales and also Filicales is doubted by some authors (Bierhorst, 1971; Kato, 1982, 1983). To determine the phylogenetic position of the Ophioglossaceae in the vascular plants, it is needed to better understand the phylogenetic relationships within the family.

The currently accepted classification of the Ophioglossaceae (e.g. Copeland, 1947; Tryon and Tryon, 1982) is mainly that of Clausen (1938) who recognized the genera *Botrychium, Helminthostachys* and *Ophioglossum* in the family. He classified *Botrychium* in three subgenera (*Osmundopteris, Sceptridium* and *Botrychium*) with seven sections, and *Ophioglossum* into the four subgenera *Ophioglossum, Rhizoglossum, Cheiroglossa* and *Ophioderma.* In his revised system, Nishida (1952) divided the Ophioglossales into two suborders, Ophioglossineae and Botrychiineae. The Ophioglossineae consisted of four genera, *Ophioglossum, Ophioderma, Cheiroglossa* and *Rhizoglossum,* belonging to the one family Ophioglossaceae, while the Botrychiineae comprised two families, Botrychiaceae with the genera *Botrychium, Sceptridium* and *Osmundopteris* (= *Botrypus*), and Helminthostachyaceae with the sole genus *Helminthostachys.* Following Clausen

Table 1. Characters in Ophioglossaceae

Code	Character	*Botrypus*	*Japanobotrychium*	*Sceptridium*	*Helminthostachys*	*Botrychium*	*Ophioglossum*
1	Rhizome	radial	radial	radial	dorsiventral	radial	radial
2	Vascular cambium	+	+	+	–	+	–
3	Leaf sheath covering	incomplete	complete	complete	complete	complete with fused slit	complete with oblique slit
4	Trophophyll size	+++	+++	++	++	+	+
5	Trophophyll dissection	++++	++++	+++	++	++	+
6	Vascular branching in leaf axis	extra-marginal	extra-marginal	extra-m./marginal	extra-m./hetero-marginal	marginal	marginal
7	Venation	free	free	free	free, parallel	free, parallel	anastomosing
8	Sporophyll position	stipe top	lower part of rachis	lower to upper part of stipe	stipe top	stipe top	stipe top (stipe base, lamina)
9	Sporophyll form	foliar	foliar	foliar	spike	foliar	spike
10	Sporophyll construction	2-dimensional	2-d.	2-d.	3-d.	2-d.	2-d.
11	Sporangium position	terminal	lateral	lateral	lateral	lateral	embedded
12	Sporangium dehiscence	longitudinal/ subtransverse	transverse	transverse	transverse	transverse	transverse
13	Suspensor	–	–	+	+	–	–

(1938) in general scheme, Kato and Sahashi (1977) recognized a fourth subgenus *Japanobotrychium* in genus *Botrychium*, which had been treated as a section of subgenus *Osmundopteris* by Clausen (1938).

The classification of Ophioglossaceae is still controversial, however. Many authors consider *Sceptridium* the most primitive, and *Botrypus* (= *Osmundopteris*) and *Botrychium sensu stricto* advanced, mainly from morphological characters (Bower, 1926; Clausen, 1938, 1954; Nishida, 1957; Nozu, 1955). Based on the vascular anatomy of leaves, Chrysler (1945) and Nishida (1952) presumed *Botrychium sens. str.* to be the most primitive, and *Botrypus* (as *Osmundopteris*) and *Sceptridium* advanced. Another different opinion on the relationship was presented by Kato (1978a) utilizing sporangia as well as such foliar characters as basal sheath, and size and cutting of leaves. *Botrypus* (as subgenus *Osmundopteris*) was regarded as the most ancestral while *Japanobotrychium, Sceptridium* and *Botrychium sens. str.* as successively specialized.

The systematic position of *Helminthostachys* is also debatable. It is generally placed as an intermediate between *Botrychium sensu lato* and *Ophioglossum* (Clausen, 1938; Nishida, 1957). Nishida (1952) recognized a monotypic Helminthostachyaceae and a family Botrychiaceae in Botrychiineae. Kato (1982) suggested a closer affinity of *Helminthostachys* to *Japanobotrychium* and *Sceptridium* among the then recognized subgenera of *Botrychium*.

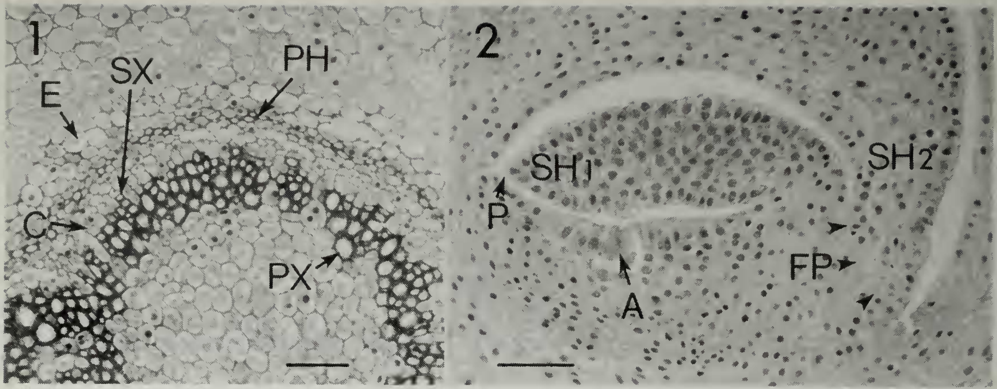
Since previous schemes of classification of Ophioglossaceae differ from each other, and none is widely accepted, the family needs re-examination from the viewpoint of phylogenetic classification. Because no closely related fossils have been found, such a systematic study must be based on comparative analysis of living genera and general paleobotanical information.

The best method of constructing a phylogenetic classification seems to me to be cladistics (e.g. Bremer and Wanntorp, 1978; Cracraft, 1974; Estabrook, 1978; Hennig, 1965, 1966; Nelson, 1974; Wagner, 1969; Wiley, 1980). In cladistics a phylogenetic classification is constructed by recognizing levels of monophyletic groups, which have synapomorphic characters derived from those of the nearest common ancestors. Monophyletic groups should be distinguished from paraphyletic (symplesiomorphic) and polyphyletic groups. Cladistics has since Hennig elaborated several different methods, each suitable for certain groups or subjects (Duncan and Stuessy, 1985; Funk and Stuessy, 1978). By adopting the basic method of cladistics (Hennig, 1965, 1966; Estabrook, 1978), an attempt is here made to examine the genealogical relationship in the Ophioglossaceae and arrange them in a phylogenetic classification. Basic monophyletic groups treated here are *Botrychium sens. str., Botrypus* (= *Osmundopteris*), *Helminthostachys, Japanobotrychium, Ophioglossum* and *Sceptridium,* although most of the genera need more detailed revisionary work.

Phylogenetic Estimation of Characters

Almost all data on the characters analyzed here are cited from previous works. The basic source for this study is Kato and Sahashi (1977) and Kato (1978a, 1978b, 1982, 1983). The characters of each genus are presented in Table 1, and character state, primitive or plesiomorphic, vs. advanced or apomorphic, is estimated in each character.

Rhizome and stele. — The rhizome is radial in construction in *Botrychium, Botrypus, Japanobotrychium, Ophioglossum* and *Sceptridium.* It has been reported that *Sceptridium* (as *Botrychium sens. lat.*) *multifidum* has a spiral phyllotaxy of (1/3)2/5(3/8) (Stevenson, 1980), *Ophioglossum petiolatum* 2/5, *O. crotalophoroides* 1/2 (Webb, 1975, 1981), and *O. pendulum* 1/3 and 3/5 (Petry, 1914). The creeping



Plates 1 & 2. *Botrychium lunaria*. Pl. 1: Transection of rhizome. *C*, cambium; *E*, endodermis; *PH*, primary phloem; *PX*, primary xylem; *SX*, secondary xylem. Pl. 2: Longisection of shoot, showing pore closing at leaf sheath. *A*, shoot apex; *FP* with arrowheads, closed pore; *P*, pore; *SH1*, leaf sheath of first leaf primordium; *SH2*, leaf sheath of second leaf primordium. Scales 100 μ.

rhizome of *Helminthostachys* is dorsiventral with two rows of leaves on the dorsal side (Farmer and Freeman, 1899). As is generally the case in vascular plants, the radial construction of rhizome in most genera of Ophioglossaceae is primitive while the dorsiventral rhizome of *Helminthostachys* is advanced.

The rhizomes of *Botrypus, Japanobotrychium* and *Sceptridium* have a cambium producing secondary vascular tissue. Although *Botrychium lunaria* has been described as having no vascular cambium (Nozu, 1956) or as having a cambium derived from the pericycle (Lang, 1913), it has the same organization as the others, excepting lower cambial activity (Takahashi, pers. commun.; Plate 1). There is no cambium in *Helminthostachys* and *Ophioglossum,* and Farmer and Freeman (1899) described the xylem as differentiating very slowly from the procambial strand in *Helminthostachys.* However, addition of xylem occurs both in *Helminthostachys* (Farmer and Freeman, 1899; Takahashi, pers. commun.) and *Ophioglossum* (Boodle, 1899). It is suggested that cambial activity has weakened and eventually disappeared, as in the evolutionary trend from a woody to herbaceous habit in Isoetales and related fossils (Rothwell and Ervin, 1985) and the flowering plants (Eames, 1961). The presence of a vascular cambium may be primitive and its absence advanced. *Helminthostachys* has a dorsiventral solenostele and *Ophioglossum* has a radial dictyostele, and this suggests a different or phylogenetically remote origin. It is interesting that Ophioglossaceae are similar to Isoetales (Lycopodiopsida) in possessing secondary tissue in very small (probably reduced) rhizomes.

Leaf sheath. — The stem apex and leaf primordia are covered by leaf sheaths to a varying extent depending on the genus (Clausen, 1938; Kato 1978a). The covering is incomplete in *Botrypus* in which sheath margins overlap along the entire length except at the base. In *Botrychium, Helminthostachys, Japanobotrychium* and *Sceptridium,* leaf sheaths cover more completely, and have a transversely elongate, small pore on the side opposite the leaf base, through which the inner cavity surrounding younger leaf primordia opens to the exterior. A further complete covering is seen in *Botrychium.* In *B. lunaria* the pore that exists in young leaf primordia disappears during leaf development (Plate 2; Kato, 1982). In *Ophioglossum* with a sunken stem apex, the pore is a narrow channel opening more or less obliquely (Holle, 1875; McAlpin, 1971). Closing of the pore takes place also in *O. termale* var. *nipponicum* (Imaichi, pers. commun.). In the light of the morphological basis that more appendicular structures are generalized, it is suggested that the

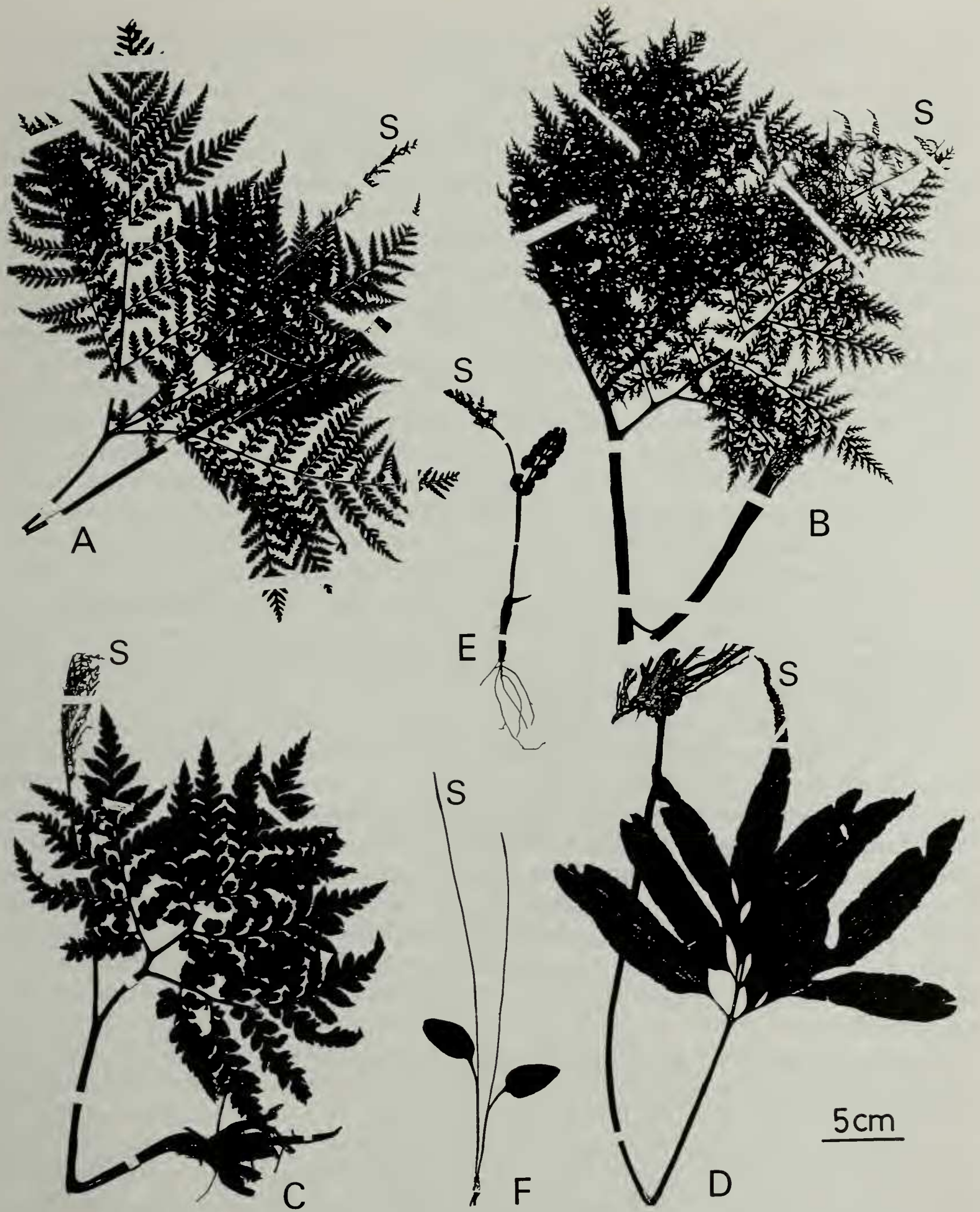


Plate 3. Silhouettes of Ophioglossaceae leaves. *A, Botrypus strictus* (*Ohba 411*, Honshu, Japan; TI); *B, Japanobotrychium lanuginosum* (*Hara et al. 6305423*, E. Nepal; TI); *C, Sceptridium daucifolium* (*Hara et al. 2458*, Darjeeling, Sikkim; TI); *D, Helminthostachys zeylanica* (*Tagawa & Iwatsuki 2170*, Ryukyus, Japan; TI); *E, Botrychium lunaria* (*Makino s.n.* 25 Jul. 1881, Mt. Fuji, Japan; TI); *F, Ophioglossum petiolatum* (*Yoshinaga s.n.* 19 Jun. 1938, Shikoku, Japan; TI); *S*, sporophyll.

incomplete covering of the leaf sheath in *Botrypus* is primitive and that of the other genera advanced. Among the latter, *Japanobotrychium*, *Sceptridium* and *Helminthostachys* are relatively primitive in this character, as compared with *Botrychium*, which has a further advanced leaf sheath.

Trophophyll morphology. — The trophophyll is variable in size and cutting in different genera, and variation in the two characters is more or less correlated (Plate 3; Clausen, 1938). *Botrypus* and *Japanobotrychium* have the largest and most dissected trophophylls in the Ophioglossaceae, and *Sceptridium* has smaller and less dissected trophophylls. The trophophyll of *Helminthostachys* is of almost the same size as that of *Sceptridium* but is much less dissected. The trophophylls of most *Botrychium* and *Ophioglossum* are the smallest, although in *Ophioglossum pendulum* they are exceptionally large (to 1 m long). They are simply pinnate in *Botrychium* while in *Ophioglossum* they are in general simple and entire, but often forked in *O. pendulum* and lobed in *O. palmatum*.

Except for *Ophioglossum pendulum* which might show some secondary leaf enlargement, Ophioglossaceae exhibit a continuous variation in leaf size and cutting between the larger and highly dissected trophophylls and the smaller and less dissected. Reduction and simplification, which is a general tendency in the lower vascular plants, is also conceivable in the Ophioglossaceae. This assumption is supported by an ontogenetic study by Imaichi and Nishida (1986) who showed apparently three-dimensional organization of trophophylls at early ontogenetic stages and the persistence of a tetrahedral apical cell at the apex of the primary through tertiary leaf axes in *Sceptridium ternatum*. It is also supported by an out-group criterion that the larger and finely dissected trophophylls are shared by *Japanobotrychium* and *Botrypus* which is an out-group for another group including the former genus, as indicated by sporangial morphology and other characters (see below). Therefore, *Sceptridium, Helminthostachys, Botrychium* and *Ophioglossum* have advanced trophophylls, compared to primitive ones in *Botrypus* and *Japanobotrychium*. Among the former, *Botrychium* has more advanced trophophylls and *Ophioglossum* has the most specialized.

Helminthostachys has a characteristic trophophyll: it is imparipinnate with oblong entire terminal and lateral pinnae, the basal and usually also the subbasal pinnae with conforming basiscopic pinnules. The presence of such basiscopic pinnules is shared with *Japanobotrychium, Sceptridium* and part of *Botrychium (B. lanceolatum)*. Moreover, *Helminthostachys* and *Sceptridium* exhibit a strong similarity in leaf shape at the young ontogenetic stages: juvenile leaves of *Helminthostachys* with three deltoid-ovate leaflets (Fig. 1) are quite similar to those of *Sceptridium japonicum* (Nishida, 1955). It is presumed that the trophophyll of *Helminthostachys* has been derived from, or from a common ancestral form of, that of *Sceptridium* which at maturity is the most similar to *Helminthostachys* in leaf size and dissection.

Vascular branching in leaf axes. — Bower (1923) recognized two types of vascular branching in fern leaves; of those he regarded marginal branching as

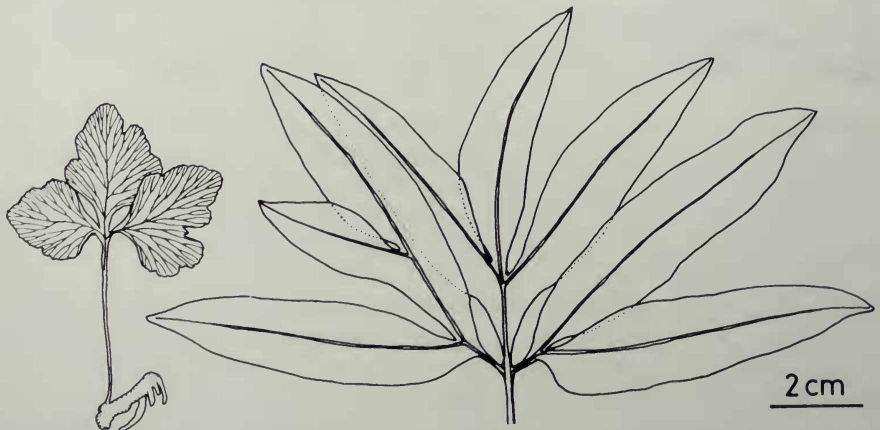


Fig. 1. Leaves of very young *(left)* and young plants of *Helminthostachys zeylanica* (*Kato et al. C-8043*, Ceram, Indonesia; TI).

primitive and extra-marginal as advanced. The same evolutionary trend was applied to the Ophioglossaceae by Chrysler (1945), Nozu (1955) and Nishida (1957). Their arguments were based on an assumption that a simpler branching type is primitive and the more elaborate, advanced, and the three-dimensional archaic branching is maintained in a simpler form in vasculature in Ophioglossaceae leaves. Kato (1978b) made a detailed re-examination of leaves of the family and criticized the validity of such an assumption. He concluded that in the Ophioglossaceae simplification from extra-marginal branching to marginal might have taken place parallel to the leaf reduction and simplification discussed above. It is estimated that in vascular branching, *Botrychium* and *Ophioglossum* are derived and other genera primitive.

It is of little doubt that marginal and hetero-marginal branchings of the sporophyll vascular bundles are each related to extra-marginal, and the first two have no direct relationship to one another (Kato, 1978b). Based on the evolutionary trend in vasculature noted above and also the phylogenetic relationship discussed below that *Botrypus* and a group of the other genera are sister-groups, the hetero-marginal branching of *Helminthostachys* and the marginal one of *Botrychium* have been derived in parallel from extra-marginal by specialization, respectively.

Venation. — Veins are free in *Botrychium, Botrypus, Helminthostachys, Japanobotrychium* and *Sceptridium*, and anastomosing in *Ophioglossum*. Elaboration from free to anastomosing venation is a general evolutionary trend in megaphyllous leaves, although the reverse tendency seems to have taken place in occasional cases, e.g. Polypodiaceae, in relation to reduction in leaf size. In Ophioglossaceae, as in general in megaphyllous plants, the anastomosing venation in *Ophioglossum* with specialized simple leaves is believed advanced, and free venation in the other genera as primitive.

Among the free-veined genera, *Botrypus, Japanobotrychium* and *Sceptridium* have pinnate and forked venation in leaf segments, while veins in *Botrychium* are parallel, and lateral veins are parallel and occasionally anastomosing in *Helminthostachys* (Bhambie and Madan, 1982). Venation of the first three genera is primitive compared with that of the last two, being parallel to the above suggested evolutionary trend in trophophyll morphology.

Position of sporophyll on trophophyll. — The sporophyll is attached at or near the top of a stipe of the trophophyll in most genera, i.e. *Botrychium, Botrypus, Helminthostachys* and most species of *Ophioglossum*. In *O. pendulum* a sporophyll occurs on the lower part of the blade which is attenuated toward the short stipe, and in *O. palmatum* several sporophylls are attached near the blade margin. *Ophioglossum bergianum* also is aberrant in having a sporophyll at the stipe base. In *Japanobotrychium* the sporophyll occurs on the lower part of the trophophyll rachis. In most species of *Sceptridium* the sporophyll is attached on the lower part of the trophophyll stipe, and in *S. daucifolium* it is at the upper part of the stipe. Thus, that sporophylls occur at or near the top of the trophophyll stipe is a general and so probably primitive character state which exists in different genera with various leaf morphology. Comparatively, *Japanobotrychium*, some species of *Ophioglossum* and *Sceptridium* are apparently advanced. They are not considered a synapomorphic group of common origin, because the position of sporophylls differs in different genera, and each primitive stage is seen in most species in *Ophioglossum*, and in *S. daucifolium* in *Sceptridium*. In *Japanobotrychium* the vascular supply to the sporophyll actually branches below the branching point of vascular supplies to the trophophyll pinnae, thus indicating a close anatomical similarity to *Botrypus* with likewise extra-marginal vascular branching (Kato, 1978b).

Form and construction of sporophyll. — The Ophioglossaceae exhibit strong leaf dimorphism, sporophylls generally consisting of lamina-lacking leaf axes and sporangia, although abnormally, a lamina develops on sporophylls to some extent, or some sporangia are produced on trophophylls. The most extraordinary are the leaves with no trophophylls in *Botrychium paradoxum* (Wagner and Wagner 1981), *Ophioglossum kawamurae* (Tagawa, 1939), and *O. lineare* (Brause, 1912). Sporophylls are foliar, variously pinnate and more or less similar to trophophylls in *Botrychium, Botrypus, Japanobotrychium* and *Sceptridium,* while they are spikes in *Helminthostachys* and *Ophioglossum.* The former is a primitive character and the latter advanced. The last two are not considered synapomorphic, because they differ considerably in form and construction. In *Helminthostachys* the sporophyll is a spike consisting of a stipe and a rachis with sporangiophores along both sides. In *Ophioglossum* it is a simple spike which is considered to consist of a stipe and a narrow blade (not a rachis), because the sporophyll has an entire margin and anastomosing veins as the trophophyll, but with areoles in few rows, necessitated by its narrowness (Fig. 2).

The sporophylls are flattened, as are leaves in general, in all the genera of Ophioglossaceae but *Helminthostachys* in which they are cylindrical: the sporangiophores are arranged in two or three irregular rows along each side of the rachis (Kato, 1978a). In the family flattened leaves are primitive and cylindrical advanced. The sporangiophores are modified branches of leaf axes and vascular branching to the sporangiophores is extra-marginal, a character common to all genera except for *Botrychium* and *Ophioglossum* (Kato, 1978a).

Position and dehiscence of sporangia. — Phylogenetic comparisons among sporangia of the vascular plants and those of Ophioglossaceae have been made by Kato (1978a, 1982, 1983). This character is the most basic for reconstructing the phylogeny in this study, and the assumed evolutionary trend is in good accordance with paleobotanical evidence and comparative morphology of the extant groups. There are two types of sporangia, each in one of two phyletic lines in the early vascular plants and their descendants. In the megaphyllous group derived from the Rhyniopsida and Trimerophytopsida, sporangia are terminal at the end of axes or veins that have been derived from axes and dehisce longitudinally from the top to the base. In the microphyllous group including the Zosterophyllopsida, sporangia are attached laterally on axes or leaves (on the adaxial side) and dehisce distally along a transverse line.

In Ophioglossaceae, belonging to the megaphyllous group, sporangia terminal at the endings of leaf axes and with longitudinal dehiscence are primitive, while those apparently lateral on the side of leaf axes (but terminal at vein-endings) and with apparently distal dehiscence are advanced (Fig. 2). The former are present in *Botrypus* (particularly *B. strictus*) and the latter in the other genera. The sporangia of *Ophioglossum,* although quite the same in morphology as those of the latter genera, are unique in being sunken and embedded in lamina tissue along the margin of the spike which is a narrow blade, and not of an axial nature, as noted above (Fig. 2). Therefore *Ophioglossum* is in a further advanced character state in the position of sporangia. It is noteworthy that a similar evolutionary trend in sporangium morphology with reference to dehiscence is seen in the leptosporangiate ferns (e.g. Bower, 1935).

Suspensor. — It has been reported that the embryo of *Helminthostachys* and *Sceptridium* has a suspensor (Bierhorst, 1971; Lang, 1914; Lyon, 1905; Nishida, 1955; Nishida and Imaichi, 1971) while no suspensor is known in the other genera. A suspensor-bearing embryo is known in various vascular plant groups such as Lycopodiales, Selaginellales, Marattiales, gymnosperms and angiosperms which have a massive or compact gametophyte (Bower, 1908; Land, 1923; Lang, 1914). Such an embryo is generally regarded as primitive. Although there is no decisive evidence for the evolutionary trend in the suspensor of Ophioglossaceae,

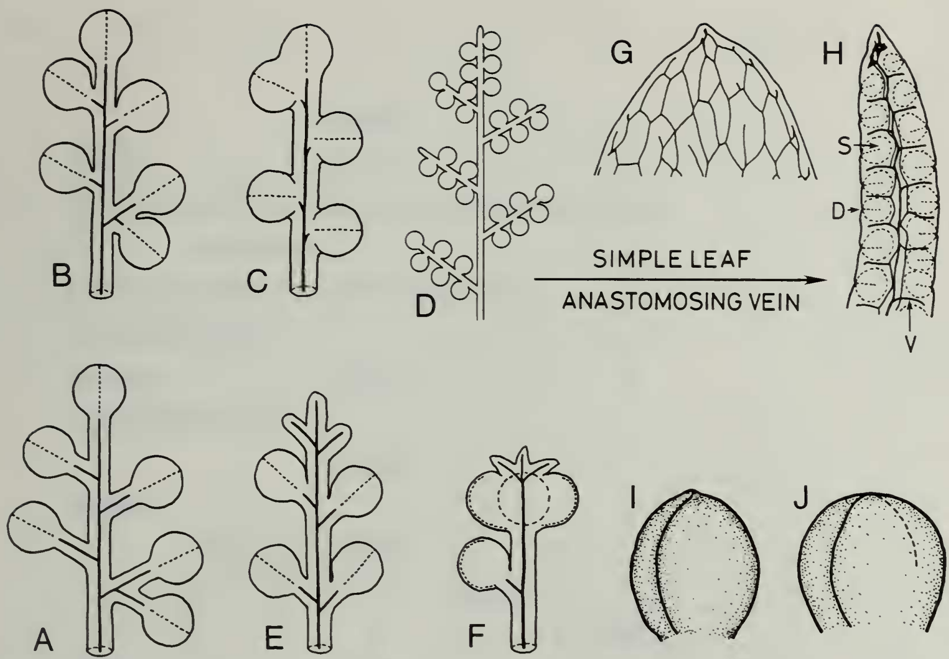


Fig. 2. Fructifications of Ophioglossaceae. *A*, presumed ancestral form; *B*, *Botrypus; C* & *D*, *Botrychium, Japanobotrychium* and *Sceptridium; E*, presumed ancestral form with distal sterile segments, equivalent to *B; F*, *Helminthostachys; G*, distal part of trophophyll of *Ophioglossum; H*, distal part of sporophyll of *Ophioglossum* (*D*, dehiscence line; *S*, sporangium; *V*, vein); *I, J*, sporangia; *I, Botrypus strictus; J, Botrychium, Japanobotrychium* and *Sceptridium.* (*A-C, E, F, I* & *J* after Kato (1978a)).

the cladogram presented below suggests the existence of a suspensor to be advanced, as far as information available at present is concerned. Another possibility is that the presence of a suspensor is ancestral in the family and this has disappeared in several genera, or that a suspensor-bearing embryo may be more widespread in the family than we know.

Phylogenetic Relationship and Classification

The evolutionary trends of all the characters examined and a deduced phylogenetic relationship between genera of Ophioglossaceae are presented in Figure 3. All character phylogenies are compatible if only we assume that the presence of a suspensor in the embryos of *Helminthostachys* and *Sceptridium* is an advanced character state, based on its absence from *Botrypus* and *Japanobotrychium* both of which are out-groups of *Helminthostachys* and *Sceptridium*, and on the embryos of *Botrychium* and *Ophioglossum* lacking a suspensor. *Botrychium* and *Ophioglossum*, and *Helminthostachys* and *Sceptridium* are each synapomorphic groups which in turn constitute a higher level synapomorphic group with *Japanobotrychium*. Among the genera, *Botrypus* is the most primitive and *Ophioglossum* is the most advanced. *Japanobotrychium*, *Sceptridium* and *Helminthostachys*, and *Botrychium* are successively more advanced genera between the two extremes.

Each genus possesses its own unique advanced character(s), showing that it is an elaboration from its nearest ancestor. Exceptionally, *Botrypus* consistently shows the primitive state of all characters examined. However, *B. cicutaria* and *B. virginianus* are derived in their sporangial morphology and *B. strictus* in its spike-like narrow sporophyll. It is, therefore, obvious that *Botrypus* is also an elaboration

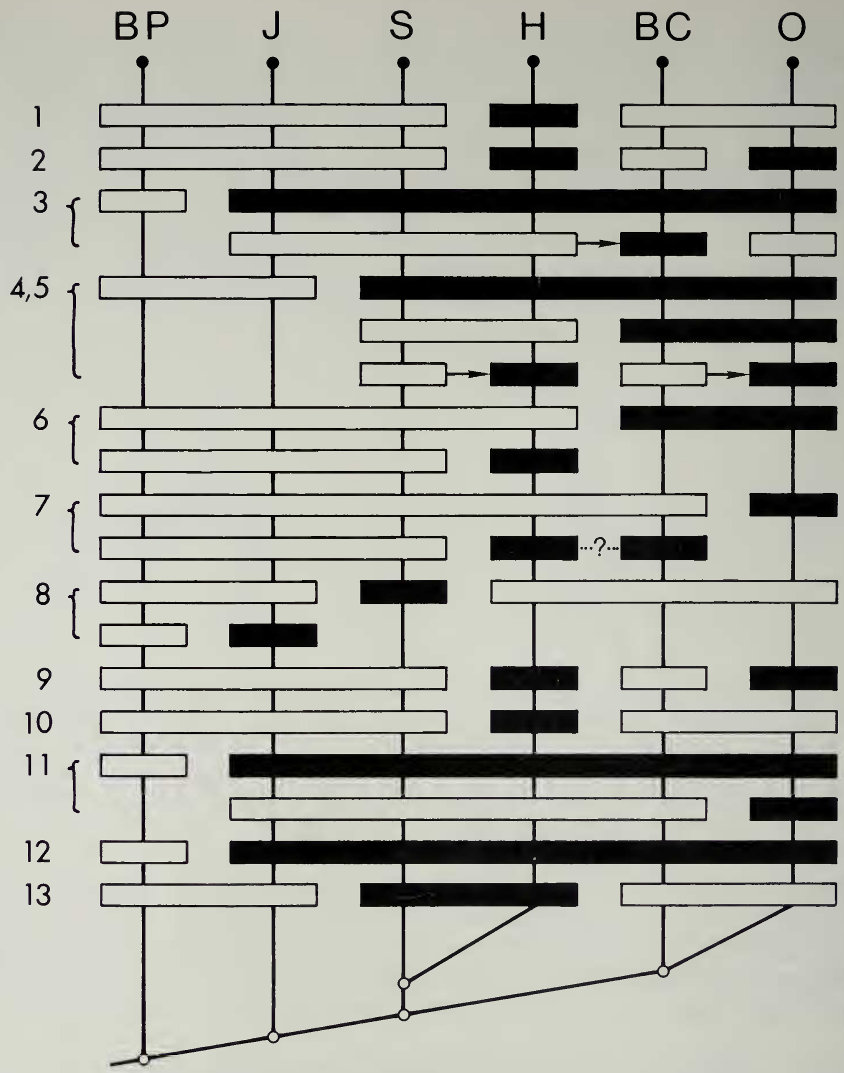


Fig. 3. Cladogram showing character state and phylogenetic relationship in Ophioglossaceae. Numbers at left side are character codes shown in Table 1. *White* and *black squares* indicate respectively primitive and advanced character states. *BP*, *Botrypus; J*, *Japanobotrychium; S*, *Sceptridium; H*, *Helminthostachys; BC*, *Botrychium; O*, *Ophioglossum*.

from the common ancestor, with its sister group being all the other extant genera of Ophioglossaceae.

The phylogenetic scheme presented in Figure 3 remarkably contradicts relationships postulated in currently accepted classifications. In these (e.g. Clausen, 1938; Nishida, 1952), *Botrychium*, *Botrypus*, *Japanobotrychium* and *Sceptridium* are united in a single group, i.e., genus *Botrychium sensu lato* or family Botrychiaceae. In the relationship obtained in this study it is not a monophyletic group and at most a paraphyletic or polyphyletic group.

For a comparison between the phylogenetic relationship of this study and that of Clausen (1938) and Nishida (1952) which differs from Clausen principally in rank of taxa, the divergence degree of the genera is estimated (Table 2). For each character, the primitive state is coded as 0 and the derived state as 0.3, 0.5, 0.7 or 1 according to the degree of deviation from the primitive state. All of the values are summed up as divergence degree. The phylogenetic relationship and divergence of

Table 2. Divergence of characters in Ophioglossaceae

Genus	Character code*												Divergence degree
	1	2	3	4	5	6	7	8	9	10	11	12	
Botrypus	0	0	0	0	0	0	0	0	0	0	0	0	0.0
Japanobotrychium	0	0	0.5	0	0	0	0	0.5	0	0	0.5	1	2.5
Sceptridium	0	0	0.5	0.5	0.3	0	0	1.0	0	0	0.5	1	3.8
Helminthostachys	1	1	0.5	0.5	0.7	0.5	0.3	0	0.5	1	0.5	1	7.5
Botrychium	0	0	1.0	1.0	0.7	1.0	0.3	0	0	0	0.5	1	5.5
Ophioglossum	0	1	0.5	1.0	1.0	1.0	1.0	0	1.0	0	1.0	1	8.5

* Following Table 1 (13 excluded).

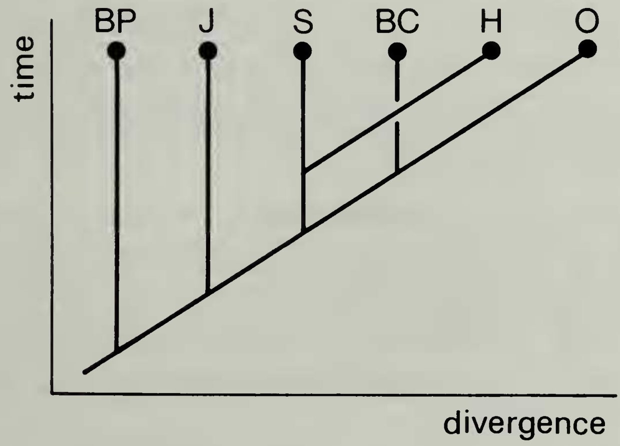


Fig. 4. Phylogenetic relationship and divergence of characters in Ophioglossaceae. *BP, Botrypus; J, Japanobotrychium; S, Sceptridium; BC, Botrychium; H, Helminthostachys; O, Ophioglossum.*

characters are shown in Figure 4. From this estimation of divergence, it is clear that *Helminthostachys* and *Ophioglossum* are the most specialized genera while *Botrychium, Botrypus, Japanobotrychium* and *Sceptridium* maintain relatively generalized characters. In the previous classifications, the former highly specialized groups have been treated as independent genera, while the latter relatively primitive genera have been classified into a single, artificial, non-monophyletic group (*Botrychium sensu lato* or Botrychiaceae).

As regards the phylogenetic classification of Ophioglossaceae, I admit that the pair of the most closely related genera, *Botrychium* and *Ophioglossum,* and *Helminthostachys* and *Sceptridium,* are distinct enough to warrant generic status, and therefore the others also deserve generic or higher rank. Although some authors (e.g. Nishida, 1952) recognize as genera *Cheiroglossa, Ophioderma* and *Rhizoglossum* as well as *Ophioglossum,* these share advanced characters as shown in Table 1 and Figure 3, so that they are best treated as infrageneric taxa. It is not feasible to classify the family into subgroups in any way although the most meaningful two groups would be *Botrypus* and then another group consisting of all the other genera. In conclusion, I propose the following classification:

Family Ophioglossaceae

1. Genus *Botrypus*
2. Genus *Japanobotrychium*
3-1. Genus *Sceptridium*
3-2. Genus *Helminthostachys*
4. Genus *Botrychium*
5. Genus *Ophioglossum*

In this scheme the genera are arranged in sequence as to reflect the phylogeny shown in Figures 3 and 4, using Cracraft's (1974) sequencing classification method. In this classification the following relationship is indicated: 1—2—3 (1—2)—4—5, and genus 1 is the most primitive and 5 the most advanced.

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